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**Pig slurry incorporation with tillage does not reduce short-term soil CO<sub>2</sub> fluxes**

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Short-communication

**Abstract**

Tillage and organic fertilization impact short-term soil CO<sub>2</sub> fluxes. However, the interactive effect of these two management practices has been rarely studied under field conditions. The objective of this study was to evaluate the impact of tillage (NT, no-tillage, and CT, conventional tillage) and fertilization strategy (PS, pig slurry, and MF, mineral fertilizer) on short-term soil CO<sub>2</sub> fluxes in a rainfed Mediterranean agroecosystem. Soil CO<sub>2</sub> fluxes were measured several times during two tillage and pre-sowing fertilization periods in 2012 and 2013 (7 and 6 times in 2012 and 2013, respectively). In the two years studied, tillage and fertilization significantly affected soil CO<sub>2</sub> fluxes, but the interaction between both factors was not significant. The application of PS resulted in a sharp and immediate increase in the soil CO<sub>2</sub> flux. One hour after the application of the organic fertilizer, soil CO<sub>2</sub> emissions increased from 0.05 to 0.70 g CO<sub>2</sub> m<sup>-2</sup> h<sup>-1</sup> and from 0.08 to 0.82 g CO<sub>2</sub> m<sup>-2</sup> h<sup>-1</sup> in 2012 and 2013, respectively. Unlike fertilization, 1 hour after tillage similar soil CO<sub>2</sub> fluxes were observed in CT and NT plots. However, after 7 h, larger fluxes were observed in CT compared with NT in both years. Cumulative CO<sub>2</sub> flux during the first 24 h after fertilization and tillage was about three- and two-fold greater in PS than in MF and in CT than in NT, respectively. The results of this study showed that in rainfed Mediterranean systems, soil management and fertilization have a noteworthy impact on short-term soil CO<sub>2</sub> losses though no interactive effects were observed between both management practices.

**Keywords:** Short-term soil CO<sub>2</sub> flux; tillage; organic fertilization; Mediterranean agroecosystems.

## Introduction

Soil stores 1500 Pg of organic carbon (C) to 1 m depth being this pool two times the C present in the atmosphere (Batjes, 1996). In agricultural soils, changes in soil organic C (SOC) levels depend on the balance between crop residues (C inputs) and microbial-derived carbon dioxide (CO<sub>2</sub>) released during the decomposition of soil organic compounds. In the absence of disturbance, the CO<sub>2</sub> produced is stored in soil matrix and gradually released to the atmosphere according to a diffusion gradient. However, under certain management practices, the CO<sub>2</sub> stored in the soil matrix can be rapidly released to the atmosphere (Rochette and Angers, 1999).

It has been demonstrated that tillage favours the short-term release of significant amounts of CO<sub>2</sub> stored in the soil structure (Reicosky et al., 1997; Rochette and Angers, 1999; Álvaro-Fuentes et al., 2007). After a tillage event, the most part of the CO<sub>2</sub> accumulated in the soil matrix can be rapidly released to the atmosphere within a few hours (Morell et al., 2010). Likewise, the addition of organic residues may also stimulate short-term soil CO<sub>2</sub> losses (Chantigny et al., 2001; Grave et al., 2015). Labile organic compounds present in animal residues may stimulate soil microbial activity triggering soil CO<sub>2</sub> fluxes just after its application (Kirchmann and Lundvall, 1993). Despite the positive response of tillage and organic fertilization on short-term soil CO<sub>2</sub> fluxes, the interactive effect of these two management practices has been rarely studied under field conditions (Grave et al., 2015).

In certain Mediterranean areas, the reduction of tillage intensity and the application of organic residues of animal origin are two promising strategies (Plaza-Bonilla et al., 2014), especially in rainfed conditions in which the reduction of operation costs is the most viable strategy to optimize farm income. According to this, our objective was to evaluate the impact of tillage (conventional tillage vs. no-tillage) and fertilization type

(mineral vs. pig slurry) on short-term soil CO<sub>2</sub> fluxes in a rainfed Mediterranean agroecosystem. We hypothesized that the interaction between tillage and fertilization type would affect short-term soil CO<sub>2</sub> losses.

## Materials and Methods

A tillage and fertilization experiment established in 2010 and located in NE Spain (41°54'12"N, 0°30'15"W) was selected for this study. The area represents the typical Mediterranean conditions with 327 mm, 1197 mm and 13.4 °C of mean annual precipitation, mean annual ETo and mean annual air temperature, respectively. The soil was classified as Typic Calcixerept (Soil Survey Staff, 2014) with the next soil characteristics at the start of the experiment (0-30 cm layer): pH (H<sub>2</sub>O, 1:2.5): 8.0; electrical conductivity (1:5): 1.04 dS m<sup>-1</sup>; organic C (g kg<sup>-1</sup>): 15.6; organic N (g kg<sup>-1</sup>): 1.4; and sand (2,000–50 µm), silt (50– 2 µm) and clay (<2 µm) content: 62, 633 and 305 g kg<sup>-1</sup>, respectively. The experiment compared two different tillage systems, conventional tillage (CT, consisting in two passes of chisel ploughing to 20 cm depth) and no-tillage (NT), together with different nitrogen fertilization strategies: three N fertilization doses (0, 75 and 150 kg N ha<sup>-1</sup>) and two types of fertilizer products (MF, mineral N and, PS, organic N with pig slurry). Both factors resulted in ten different tillage-fertilization treatments replicated three times. Plot size was 40 x 12 m in the organic N fertilization treatment and 40 x 6 m in the mineral N fertilization treatment. For this study, only four treatments were selected: NT mineral fertilized with 150 kg N ha<sup>-1</sup> (NT-MF); NT fertilized with 150 kg N ha<sup>-1</sup> of pig slurry (NT-PS); CT mineral fertilized with 150 kg N ha<sup>-1</sup> (CT-MF); and CT fertilized with 150 kg N ha<sup>-1</sup> of pig slurry (CT-PS). The cropping system consisted of a barley (*Hordeum vulgare* L.) monoculture and the application of 150 kg N ha<sup>-1</sup> of fertilizer in split half at planting

and half at the beginning of tillering. The composition of the PS fertilizer did not differ between years, with 56 and 54 g kg<sup>-1</sup> of dry matter, 24.2 and 23.7 g kg<sup>-1</sup> of Kjeldahl N and 36.4 and 36.4 g kg<sup>-1</sup> of ammonium N for the 2012-2013 and 2013-2014 seasons, respectively.

Soil CO<sub>2</sub> flux measurements were performed during the tillage and pre-sowing fertilization operations of the 2012-2013 (referred as 2012) and 2013-2014 (referred as 2013) cropping seasons. Fertilization and tillage operations were performed the same day with only 30 minutes difference. In the 2012 and 2013 cropping seasons, tillage and fertilization operations were performed on 20 November 2012 and 1 November 2013, respectively. Soil CO<sub>2</sub> flux was measured with an open chamber system (model CFX-1, PPSystems, Hertfordshire, London) connected to an infrared gas analyser (model EGM-4, PPSystems, Hertfordshire, London). The chamber had a cylindrical diameter of 21 cm, covering a soil surface of 346 cm<sup>2</sup>, and it was directly inserted 2 cm deep in the soil to prevent gas leak to the atmosphere. The air flow rate of the chamber was adjusted to 900 mL min<sup>-1</sup>. Two gas observations per plot were taken. Both years, soil CO<sub>2</sub> flux measurements were taken 24 h prior to tillage and fertilization operations and 1 h, 7 h and 24 h after operations. Additionally, in 2012, measurements were also taken 48 h, 72 h and 168 h after operations. In 2013, additional measurements were also performed 96 h and 144 h after operations. In both years, the time prior or after operations was taken considering the time of fertilization as the reference, despite the 30 minutes delay between fertilization and tillage. Cumulative emissions during the first 24 h after operations were calculated from linear interpolation between consecutive samplings using the trapezoid rule (Morell et al., 2010).

For each year, soil CO<sub>2</sub> fluxes were analysed performing a repeated measures analysis of variance (ANOVA) using the nlme package (Pinheiro et al., 2014) of the R software

version 3.0.2. Due to lack of normality (tested with the Shapiro-Wilks test), CO<sub>2</sub> flux data was transformed using a Box-Cox procedure with the *cran* package (Fox and Weisberg, 2011) of the R software. Cumulative fluxes were analyzed with ANOVA analysis considering tillage, fertilization and year as main factors.

## Results and Discussion

In the two years studied, tillage and fertilization affected soil CO<sub>2</sub> fluxes, but the interaction between both factors was not significant ( $P < 0.05$ ). Consequently, the main hypothesis of the study was rejected. The application of PS resulted in a sharp and immediate increase in the soil CO<sub>2</sub> flux. According to Fig. 1, 1 hour after the application of the organic fertilizer, soil CO<sub>2</sub> emissions increased from 0.05 to 0.70 g CO<sub>2</sub> m<sup>-2</sup> h<sup>-1</sup> and from 0.08 to 0.82 g CO<sub>2</sub> m<sup>-2</sup> h<sup>-1</sup> in 2012 and 2013, respectively. However, the application of MF did not increase soil CO<sub>2</sub> fluxes. The increase in soil CO<sub>2</sub> fluxes after PS addition was also observed in other similar field experiments performed in different agroecosystems (Chantigny et al., 2001; Grave et al., 2015). Kirchmann and Lundvall (1993) concluded that, once in the soil, the fatty acids present in the PS are fast mineralized with the concomitant flux in CO<sub>2</sub>. Another possible explanation could be related to the rapid release of carbonates accumulated during anaerobic storage of PS (Chantigny et al., 2001). However, this release is only expected in acidic soils when carbonates are decomposed to CO<sub>2</sub> and not in high pH soils like the soil of our experimental site with a pH higher than 8.

In Mediterranean semiarid conditions, microbial activity is constrained by soil moisture (Almagro et al., 2009). Furthermore, soil wetting after drought periods results in an increase in soil CO<sub>2</sub> emissions from the recovery and activation of microbial activity (Morell et al., 2010). In our experiment, total slurry applied was on average 21 and 20

$\text{m}^3 \text{ ha}^{-1}$  in 2012 and 2013, respectively. This volume of slurry led to an increase in surface soil water content (5 top cm) from 19% to 25% and from 15% to 22% in 2012 and 2013, respectively (comparing values 24 h before and 1 h after fertilization) (data not shown). The increase in soil water content in the first cm of the soil due to the addition of PS could also stimulate the flux of soil  $\text{CO}_2$  to the atmosphere. The positive effect of the labile C compounds present in the PS on microbial activity could be enhanced by wetter soil conditions after the addition of PS. At the same time, rewetting of dry soils stimulates short-term  $\text{CO}_2$  flux (Fierer and Schimel, 2002; Morell et al., 2010). Different processes have been associated to the increase of soil  $\text{CO}_2$  after rewetting: (i) rapid mineralization of labile substrates accumulated during the dry period; (ii) soil aggregate breakdown and mineralization of occluded organic matter released; and (iii) physical displacement out to the atmosphere of the  $\text{CO}_2$  stored in the soil structure (Kim et al., 2012).

Unlike fertilization, 1 hour after tillage similar soil  $\text{CO}_2$  fluxes were observed in CT and NT plots (Fig. 1). However, after 7 h, larger fluxes were observed in CT compared with NT in both years. This last difference was especially significant in 2012 when mean soil  $\text{CO}_2$  fluxes were  $0.49$  and  $0.21 \text{ g CO}_2 \text{ m}^{-2} \text{ h}^{-1}$  in CT and NT plots, respectively (Fig. 1). Indeed, in 2012, soil  $\text{CO}_2$  fluxes 7 h after tillage were even higher than after 1 h, contrary to previous findings obtained in similar rainfed Mediterranean conditions in which the highest fluxes were always obtained in the nearest sampling time after tillage (Álvaro-Fuentes et al., 2007; Morell et al., 2010). It is widely accepted that the loss of the  $\text{CO}_2$  entrapped within soil structure due to changes in soil physical conditions explains the peak of this gas after tillage (Reicosky and Lindstrom, 1993; Rochette and Angers, 1999). The changes in soil structure after tillage caused a rapid burst of soil  $\text{CO}_2$  which is expected immediately after tillage operations (Reicosky et al., 1997).



Accordingly, the CO<sub>2</sub> peak observed 7 h after tillage in 2012 may not be associated to a physical release of gas entrapped in soil structure but to a positive synergistic effect of CT on short-term microbial activity after PS addition. Increases in soil aeration after tillage could stimulate microbial activity contributing to the mineralization of labile C compounds in the PS.

Short-term CO<sub>2</sub> loss induced by fertilization and tillage occurred within the first 24 h (Fig. 1). Accordingly, cumulative fluxes during this time interval were calculated and differences among treatments further analysed. As observed for hourly fluxes, tillage and fertilization interaction was neither significant. As observed in Fig. 2, in both years, cumulative CO<sub>2</sub> flux during the first 24 h after fertilization was about three-fold greater in PS than in MF. Cumulative losses between tillage systems were lower than between fertilization strategies with almost two-fold greater cumulative CO<sub>2</sub> flux in CT compared with NT (Fig. 2). Despite these differences, it is important to highlight that the increase in CO<sub>2</sub> flux measured in the treatments with PS during the first 24 h after tillage represented only a small part of the total organic C added with the PS. For example, in 2012, the mean cumulative CO<sub>2</sub> loss in the PS treatment was 100 kg CO<sub>2</sub> ha<sup>-1</sup>, which represented less than a tenth of the estimated total C applied with the pig slurry (27 kg C ha<sup>-1</sup> vs. 468 kg C ha<sup>-1</sup>, emitted and applied, respectively).

The mean cumulative soil CO<sub>2</sub> loss during the first 24 h after tillage was significantly different ( $P < 0.05$ ) between years. While in 2012 the mean cumulative flux was 68 kg CO<sub>2</sub> ha<sup>-1</sup>, in 2013 this value dropped to 46 kg CO<sub>2</sub> ha<sup>-1</sup>. Measured soil water content to 60 cm depth was about 15% higher in October 2012 than in October 2013 (110 vs. 95 mm, respectively) (data not shown). The wetter conditions found during the weeks previous to tillage and fertilization in 2012 could have stimulated microbial activity

facilitating the accumulation of CO<sub>2</sub> in the soil matrix that was afterwards released to the atmosphere.

According to our experiment, the fertilization with PS does not decrease soil CO<sub>2</sub> fluxes. Indeed, when PS is added together with CT implementation, it may have a double effect on short-term soil CO<sub>2</sub> fluxes. Firstly, CT may loose the soil facilitating physical release of CO<sub>2</sub> entrapped in the soil structure and, secondly, it may stimulate microorganisms responsible for the decomposition of easily degraded organic compounds present in the PS. According to our study, the difference of time between both phases would be small and occurring within the first 24 hours.

The results of this study showed that in rainfed Mediterranean systems, soil tillage and fertilization have a noteworthy impact on short-term soil CO<sub>2</sub> losses though no interactive effects were observed between both management practices. Despite the importance of PS in terms of soil CO<sub>2</sub> flux increase during the first 24 h after fertilization, the cumulative C loss in the short-term is small compared with the total organic C applied. Based on the results obtained in this study, we recommend further research to evaluate the short-term impact of other organic fertilizers and the possible interaction with other tillage implements on soil C dynamics and CO<sub>2</sub> fluxes.

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**Figure captions**

Figure 1. Short-term soil CO<sub>2</sub> flux following tillage (CT, conventional tillage; NT, no-tillage) and fertilization (MF, mineral fertilization; PS, pig slurry) in 2012 and 2013. For a given sampling time, asterisks indicate significant differences between factor levels ( $P<0.05$ ).

Figure 2. Cumulative soil CO<sub>2</sub> losses during the first 24 hours after tillage (CT, conventional tillage; NT, no-tillage) and fertilization (MF, mineral fertilization; PS, pig slurry) in 2012 and 2013. For a given year and main factor (tillage or fertilization), different lowercase letters indicate significant differences among treatments ( $P<0.05$ ).

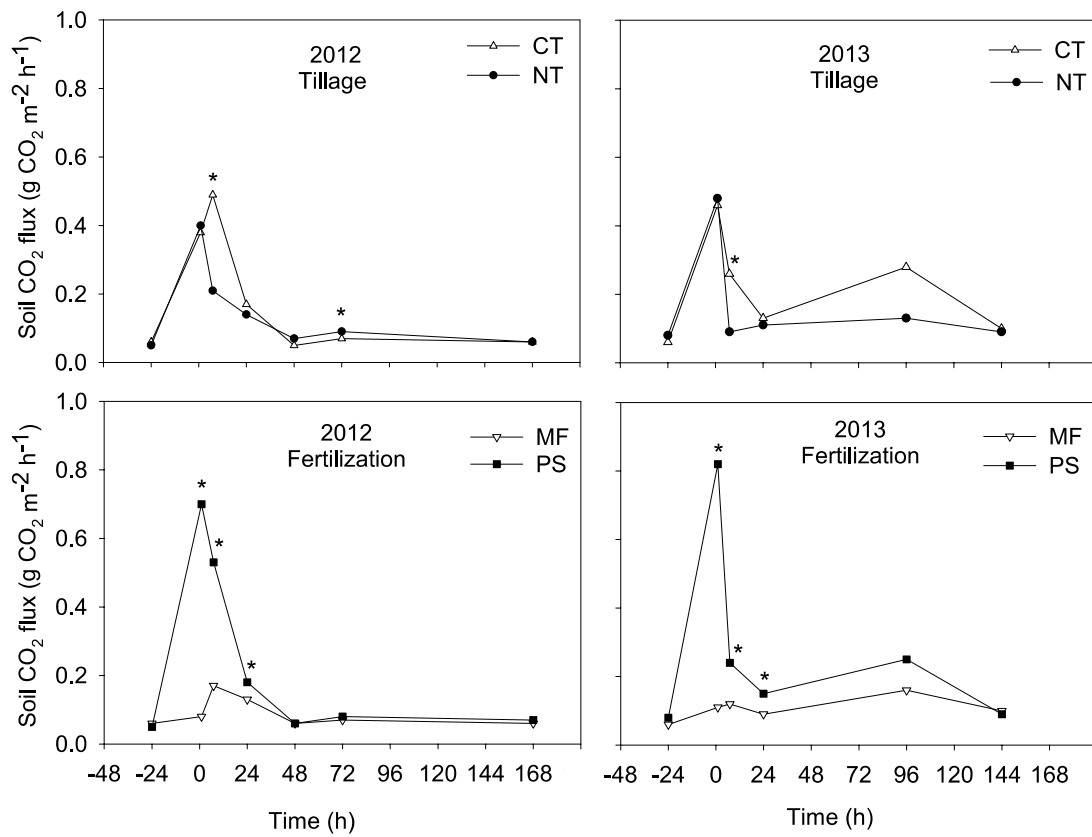


Fig. 1.

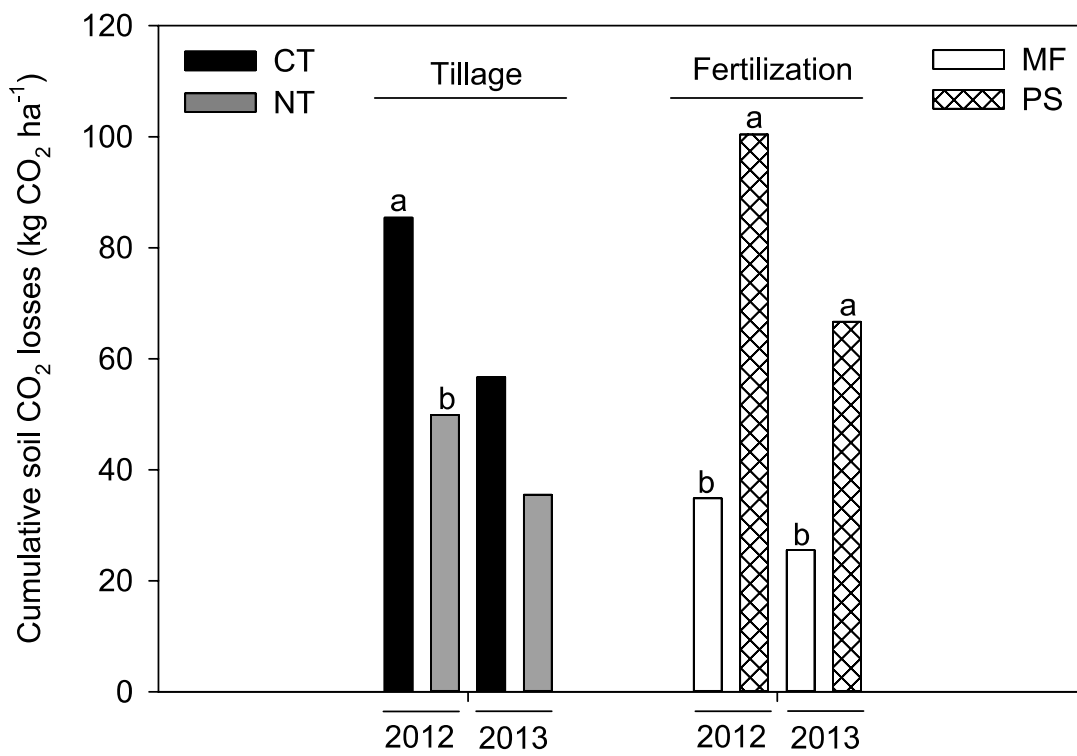


Fig. 2.

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